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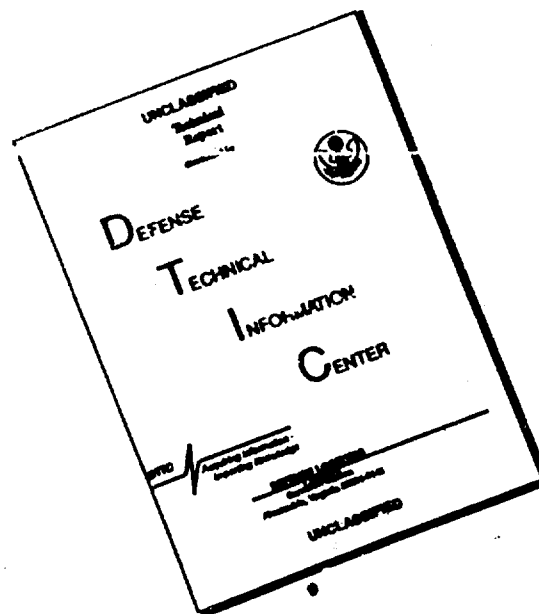
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# 2318

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Dissemination of Cl. Botulinum and Cl. Tetani in the Soil of Some  
Regions in the Armenian SSR

By: L.M. SHISHULINA

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(Translated by : Edward Lachowicz, Maryland, Medical-Legal Foundation, Inc., 700 Fleet Street, Baltimore, Maryland)

On the basis of conducted soil tests and examinations of the gastrointestinal tract contents of fishes, A.T. KRAVCHENKO et al. (1960) and L.M. SHISHULINA (1961) reported a general dissemination of Cl. botulinum throughout the territory of the USSR. The greatest dissemination of the causative agent of botulism was observed in the soil of southern regions of the country (northern CAUCASUS, PERGANA) and in the silt-covered shores of some lakes and river basins (western shores of the CASPIAN SEA, AZOV SEA and DON River delta). Thus, in the regions that have a high percentage of the soil infection by botulism bacterium there is a potential danger of flareups as well as of sporadic cases of botulism. Our attention was drawn to the fact that in recent years botulism cases were reported in the Armenian SSR that resulted from eating canned (common) purslane; its production began on a small scale at the end of 1949. Then, as the canning advanced to a large scale output in 1953, first instances of poisonings appeared. The cause was the inferior condition of canned food due to incomplete removal of the soil and sand particles from the original staples (A.V. ZAKARYAN

et al., 1960). Thus, it was expedient to ascertain a dissemination degree of *Cl. botulinum* in the soil of various regions in the Armenian SSR.

We found no indicants in the literature as to any dissemination of *Cl. tetani* on the territory of this republic. Yet, on the basis of high incidence of tetanus in the Armenian SSR (1.4 cases per 100,000 population), K.I. MATVEEV (1960) included this republic in the first zone, i.e. in the zone with the highest coefficient of the disease rate (1.3 to 5.7 per 100,000 population). These findings provided a basis for assumption that there is a considerable dissemination of *Cl. tetani* in the soil and in the surrounding media of the Armenian SSR. In order to clarify the conditions, we conducted soil tests in some regions of the republic. The investigation technique has been described in reports of A.T. KRAVCHENKO, L.M. SHISHULINA, G.P. GALKINA and L.V. GORETSKA (1960). Each sample of the soil was suspended in 1% peptone water. One part of the suspension was inoculated on the KITT-TAROCCHI culture medium, following a prior heating in water bath at 80° for 30 minutes and after cultivation period at 34°; the other part was inoculated without a prior heating but following incubation at 25°. The activity of each inoculation was tested on the 5th and 14th day of its growth by means of intraperitoneal administration to 2 white mice a dose of 0.5 ml of the culture fluid in a concentration of 1:2.

The determination of all active specimens was carried out by the neutralization method of the toxin by using polyvalent antitoxin and antitetanic serums and, then, by monovalent antitoxin serums.

*Cl. botulinum* was detected in 9 specimens (14.3%) and *Cl. tetani* in 18 specimens (28.5%). We observed a diversified dissemination of the causative agent of botulism and that of tetanus in the soil of various examined localities. We detected *Cl. tetani* in 7 out of 8 specimens in the soil used for vineyards, but none in the soil taken from the RAZDAN River shore; then, in the vicinity of the SEVAN Lake we detected tetanic rod in 2 specimens of the soil out 10 examined samples. The degree of the soil infection by tetanus was approximately the same in other localities. Among the examined sections the highest dissemination degree of the causative agent of botulism was found in the soil along the shore of the CEDAR River, in the vicinity of the collective farm's market place of YEREVAN, where, out of 9 examined specimens, we detected 5 positive samples. The shore of the CEDAR River has been polluted by waste matter and this, perhaps, contributed to the preservation and breeding of *Cl. botulinum*. In contrast with this, we detected only 2 samples containing *Cl. botulinum* out of 14 examined soil specimens taken from the shores locality of the mountain river RAZDAN where it passes close to the outskirts of the town. Here, *Cl. botulinum* was found in the soil taken from the mountain slope located about 100 m from the SEVAN Lake.

Our findings revealed that in the soil of the Armenian SSR can be found the causative agent of botulism of the types A, B, C and E, also that most specimens contained *Cl. botulinum* type A (5 samples contained *Cl. botulinum* type A, 2 samples - type B, 1 sample - types A, B, C and 1 sample - type E). We stated in previous reports that, comparing the dissemination of *Cl. botulinum* in the

soil throughout the Soviet Union and in individual regions, type B is considered predominant. Yet, the difference in dissemination of various types of the causative agent of botulism speaks for itself, namely that each region has its own peculiarity in dissemination of Cl. botulinum in the soil.

We should also add that most specimens containing Cl. botulinum were not heated prior to inoculation, but were allowed to incubate at 25° (see Table 1). We found that the same samples used for in-

Table 1

Dissemination of Cl. Botulinum Contingent upon the Method of Specimens' Processing

Cultivation temperature	Processing of material prior to inoculation	Quantity of specimens containing Cl. botulinum			
		A	B	AC	E
25	Without heating	5 <sup>1)</sup>	3	--	1
34	Heating at 80° for 30 minutes	1	-	1 <sup>2)</sup>	-

1) - One sample contained Cl. botulinum in a preheated and in not heated suspension of the soil.

2) - Cl. botulinum type B was detected in this not heated specimen cultivated at 25°.

oculation after heating became inactive, with the exception of one specimen which produced a "nonspecific" activity, namely the progress of the disease run a nontypical course in white mice with regard to the botulism intoxication and negative reaction following the neutralization of toxin by polyvalent antitoxin serum. Thus, for more specific findings on dissemination of Cl. botulinum in the examined material we recommend that inoculations should include heated as

well as not heated material.

The absence of *Cl. botulinum* in inoculations of samples heated and then cultivated at 34° can, perhaps, be explained either by the thermolability of spores, or by their negligible quantities among the available vegetative <sup>microbial</sup> forms. The latter assumption tends to indicate a breeding of *Cl. botulinum* in the soil.

Data obtained by us and the presence of botulism cases that resulted from eating canned purslane confirm again the necessity of taking into account the semination of *Cl. botulinum* in moist grounds and in their environments; this should be particularly considered at the time when a new cannery is organized, or new canned foods are processed.

The high percentage of dissemination of *Cl. tetani* in the soil confirms the necessity of carrying out preventive measures proposed by K.I. MATVEEV.

#### Conclusions

1. We detected *Cl. botulinum* in 9 specimens (14.3%) and *Cl. tetani* in 18 specimens (28.5%) after examination of 63 samples of soil.

2. The soil in the Armenian SSR contains *Cl. botulinum* of the A, B, C and E types. The highest dissemination is that of *Cl. botulinum* type A. Out of 9 specimens containing *Cl. botulinum*, 5 were obtained in the vicinity of the collective farm's market place (GERDAR River shore) of YEREVAN.

3. In most cases *Cl. botulinum* was detected after inoculations of soil samples not heated previously; perhaps, this can be explained by the thermolability of spores, or by the availability of vegetative

microorganic forms.

4. Emlosures of soil used for a vineyard and orchard revealed the highest dissemination of Cl. tetani.

Literature Cited

ZAKARYAN, A.V. Proceedings of the 1st Congress of Hygienists, Epidemiologists, Microbiologists and Infectionists in the Armenian SSR. YEREVAN, 1960, p.141. - KRAVCHENKO, A.T., SHISHULINA, L.M., GALKINA, G.P. et al. in book: Anaerobic infections. Moscow 1960, p.268. - MATVEEV, K.I. Epidemiology and prophylaxis of tetanus. Moscow 1960.